

**Amendments to the Claims:**

The following Listing of Claims replaces all prior versions and listings of the claims in this application.

**Listing of the Claims**

1 (Currently Amended): A method for determination of an analyte in a sample in a flow matrix by use of a transport flow of one or more biospecific affinity reactants, at least one of which is analytically detectable (Reactant\*) and one of which is firmly anchored in the matrix (Reactant I), and the flow matrix comprises:

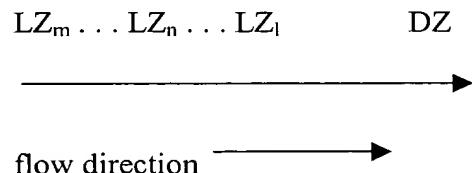
A) an application zone adapted for application of liquid (LZ), which liquid contains buffer and sample and optionally reactants needed for a complete determination, but not Reactant I,

B) a detection zone (DZ) with the firmly anchored reactant (Reactant I) located downstream of LZ, and

C) optionally one or more zones in which any of the reactants needed for a complete determination, but not Reactant I, has been pre-deposited,

wherein (i) the flow towards the detection zone is initiated by addition of the liquid with sample in the application zone LZ for transport of analyte and reactants towards the detection zone (DZ), and (ii) the amount of the Reactant\* bound to DZ is detected, wherein the detected amount is correlated to the amount of analyte in the sample, wherein

I. the flow matrix comprises at least two application zones for liquid LZ arranged substantially adjacent to each other:



wherein

- a)  $LZ_n$  is an application zone for liquid, and  $n$  is the position of the application zone  $LZ_n$ ,
- b)  $m$  is the total number of application zones in which flow is initiated,  $m$  is greater than or equal to 2, and  $m$  is not equal to  $n$ , wherein  $LZ_m$  is the farthest upstream liquid application zone,
- c) one  $LZ_n$  is an application zone for sample ( $LZ_n\text{-S}$ ) and one  $LZ_n$  is for Reactant\* ( $LZ_n\text{-R}^*$ ) with  $n'' \geq n'$ ;
- d)  $\longrightarrow$  is the direction of the flow, and
- e) DZ is the detection zone, and

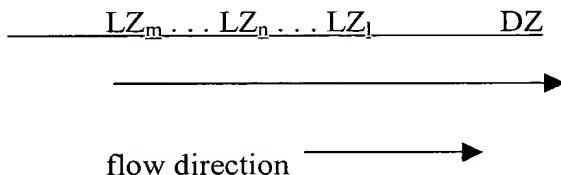
II. flow is initiated by adding liquid to each zone  $LZ_m \dots LZ_n \dots LZ_1$  in such a way that liquid <sub>$n+1$</sub> , added to the application zone  $LZ_{n+1}$ , contacts the flow matrix substantially simultaneously with and is transported through the matrix immediately after liquid <sub>$n$</sub>  added to the nearest downstream application zone  $LZ_n$ .

2 (Currently Amended): A The method according to claim 1, wherein for determination of an analyte in a sample in a flow matrix by use of a transport flow of one or more biospecific affinity reactants, at least one of which is analytically detectable (Reactant\*) and one of which is firmly anchored in the matrix (Reactant I), and the flow matrix comprises:

- A) an application zone for application of liquid (LZ), which liquid contains buffer and sample and optionally reactants needed for a complete determination, but not Reactant I,
- B) a detection zone (DZ) with the firmly anchored reactant (Reactant I) located downstream of LZ, and

C) optionally one or more zones in which any of the reactants needed for a complete determination, but not Reactant I, has been pre-deposited,  
wherein (i) the flow towards the detection zone is initiated by addition of the liquid with sample in the application zone LZ for transport of analyte and reactants towards the detection zone (DZ), and (ii) the amount of the Reactant\* bound to DZ is detected, wherein the detected amount is correlated to the amount of analyte in the sample, wherein

I. the flow matrix comprises at least two application zones for liquid LZ arranged substantially adjacent to each other:



wherein

- a) LZ<sub>n</sub> is an application zone for liquid, and n is the position of the application zone LZ<sub>n</sub>,
- b) m is the total number of application zones in which flow is initiated, m is greater than or equal to 2, and m is not equal to n, wherein LZ<sub>m</sub> is the farthest upstream liquid application zone,
- c) one LZ<sub>n</sub> is an application zone for sample (LZ<sub>n</sub>S) and one LZ<sub>n</sub> is for Reactant\* (LZ<sub>n</sub>R\*) with n'' > n';
- d) → is the direction of the flow, and
- e) DZ is the detection zone, and

II. flow is initiated by adding liquid to each zone LZ<sub>m</sub> . . . LZ<sub>n</sub> . . . LZ<sub>1</sub> in such a way that liquid<sub>n+1</sub>, added to the application zone LZ<sub>n+1</sub>, contacts the flow matrix substantially

simultaneously with and is transported through the matrix immediately after liquid<sub>n</sub> added to the nearest downstream application zone LZ<sub>n</sub>.

3 (Previously Presented): The method according to claim 1, wherein n" = n'.

4 (Previously Presented): The method according to claim 1, wherein Reactant\* is pre-deposited in its application zone (LZ<sub>n</sub>\*R\*).

5 (Cancelled).

6 (Previously Presented): The method according to claim 1, wherein LZ<sub>n+1</sub> is upstream and immediately adjacent.

7 (Previously Presented): The method according to claim 1, wherein application of liquid is performed simultaneously in all LZ<sub>m</sub> . . . LZ<sub>n</sub> . . . LZ<sub>1</sub>.

8 (Previously Presented): The method according to claim 1, wherein 2 ≤ m ≤ 6; n' is 1, 2 or 3, n" > n'; LZ<sub>n'+1</sub>, LZ<sub>n'+2</sub>, LZ<sub>n'+3</sub>, LZ<sub>n'-1</sub>, and LZ<sub>n'-2</sub> are application zones for liquids intended for transport of Reactant\* or other reactant or buffer without reactant.

9 (Previously Presented): The method according to claim 1, wherein at least one of the zones LZ<sub>m</sub> . . . LZ<sub>n</sub> . . . LZ<sub>1</sub> comprises a pad or material layer applied on the flow matrix.

10 (Previously Presented): The method according to claim 1, wherein the zones LZ<sub>m</sub> . . LZ<sub>n</sub> . . LZ<sub>1</sub> have zone spacers between each other.

11 (Previously Presented): The method according to claim 1, wherein a composition of liquid flow from an application zone LZ<sub>n</sub> is not the same as a composition of liquid flow from the nearest adjacent application zone LZ in which flow is initiated.

12 (Previously Presented): The method according to claim 1, wherein at least one reactant, other than Reactant\*, is pre-deposited in an application zone LZ<sub>n...R</sub> for liquid intended for transport of the reactant.

13 (Previously Presented): The method according to claim 1, wherein 2 ≤ m ≤ 6 and n' for the application zone for sample (LZ<sub>n</sub>S) is 1, 2 or 3.

14 (Previously Presented): The method according to claim 1, wherein Reactant\* has biospecific affinity for the analyte so that Reactant\* is incorporated into a complex Reactant'- --Analyte---Reactant\* in the detection zone in an amount related to the amount of analyte in the sample, in which complex Reactant' has biospecific affinity to the analyte and is

- (a) Reactant I, or
- (b) a reactant to which Reactant I exhibits biospecific affinity and which is transported from LZ<sub>n</sub>S or from an application zone downstream of LZ<sub>n</sub>S.

15 (Previously Presented): The method according to claim 1, wherein the matrix comprises at least one calibrator zone (CZ), in which calibrator is bound to, or in advance has been bound to the matrix.

16 (Previously Presented): The method according to claim 15, wherein the calibrator zone or zones (CZ) have a binder for the calibrator firmly anchored in the matrix, the calibrator optionally being pre-deposited in the matrix upstream of the calibrator zone or zones.

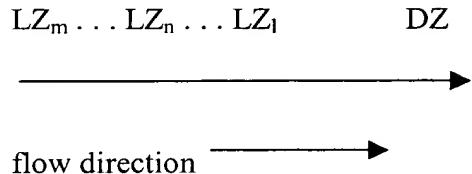
17 (Previously Presented): The method according to claim 1, wherein the method is performed as part of diagnosing allergy or autoimmune disease.

18 (Previously Presented): A device for determination of an analyte in a sample in a flow matrix by use of a transport flow of one or more biospecific affinity reactants, at least one of which is analytically detectable (Reactant\*) and one of which is firmly anchored in the matrix (Reactant I), said device comprising a flow matrix having:

- A) an application zone for liquid (LZ) containing buffer and sample and optionally reactants needed for a complete determination, but not Reactant I,
- B) a detection zone (DZ) with the firmly anchored reactant (Reactant I) located downstream of LZ, and
- C) optionally one or more zones in which any of the reactants has been pre-deposited,

wherein

the flow matrix comprises at least two application zones for liquid arranged substantially adjacent to each other:



wherein

- a)  $LZ_n$  is an application zone for liquid, and  $n$  is the position of the application zone  $LZ_n$ ,
- b)  $m$  is the total number of application zones in which flow is initiated,  $m$  is greater than or equal to 2, and  $m$  is not equal to  $n$ , wherein  $LZ_m$  is the farthest upstream liquid application zone,
- c) one  $LZ_n$  is an application zone for sample ( $LZ_n\text{-S}$ ) and one  $LZ_n$  is for Reactant\* ( $LZ_n\text{-R}^*$ ) with  $n'' \geq n'$ ;
- d)  $\longrightarrow$  is the direction of the flow, and
- e)  $DZ$  is the detection zone, wherein the device is adapted, when flow is initiated by adding liquid to each zone  $LZ_m \dots LZ_n \dots LZ_1$  in such a way that liquid <sub>$n+1$</sub>  added to the application zone  $LZ_{n+1}$ , contacts the flow matrix substantially simultaneously with and is transported through the matrix immediately after liquid <sub>$n$</sub> , added to the nearest downstream application zone  $LZ_n$ .

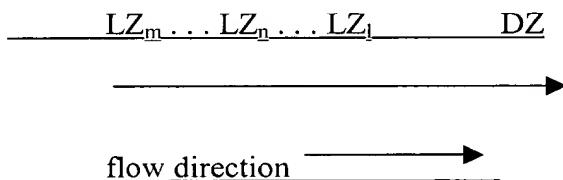
19 (Currently Amended): A The device according to claim 18 for determination of an analyte in a sample in a flow matrix by use of a transport flow of one or more biospecific

affinity reactants, at least one of which is analytically detectable (Reactant\*) and one of which is firmly anchored in the matrix (Reactant I), said device comprising a flow matrix having:

- A) an application zone for liquid (LZ) containing buffer and sample and optionally reactants needed for a complete determination, but not Reactant I,
- B) a detection zone (DZ) with the firmly anchored reactant (Reactant I) located downstream of LZ, and
- C) optionally one or more zones in which any of the reactants has been pre-deposited,

wherein

the flow matrix comprises at least two application zones for liquid arranged substantially adjacent to each other:



wherein

- a) LZ<sub>n</sub> is an application zone for liquid, and n is the position of the application zone LZ<sub>n</sub>,
- b) m is the total number of application zones in which flow is initiated, m is greater than or equal to 2, and m is not equal to n, wherein LZ<sub>m</sub> is the farthest upstream liquid application zone,
- c) one LZ<sub>n</sub> is an application zone for sample (LZ<sub>n</sub>·S) and one LZ<sub>n'</sub> is for Reactant\* (LZ<sub>n'</sub>·R\*), wherein n'' > n' and the device is intended for sequential transport of analyte and Reactant\*;

d) is the direction of the flow, and

e) DZ is the detection zone, wherein the device is adapted, when flow is initiated by adding liquid to each zone LZ<sub>m</sub> . . . LZ<sub>n</sub> . . . LZ<sub>1</sub> in such a way that liquid<sub>n+1</sub> added to the application zone LZ<sub>n+1</sub>, contacts the flow matrix substantially simultaneously with and is transported through the matrix immediately after liquid<sub>n</sub>, added to the nearest downstream application zone LZ<sub>n</sub>.

20 (Previously Presented): The device according to claim 18, wherein n" = n' and the device is intended for simultaneous transport of analyte and Reactant\*.

21 (Previously Presented): The device according to claim 18, wherein Reactant\* is pre-deposited in its application zone (LZ<sub>n"</sub>R\*).

22 (Previously Presented): The device according to claim 18, wherein LZ<sub>n+1</sub> is upstream and immediately adjacent.

23 (Previously Presented): The device according to claim 18, wherein 2 ≤ m ≤ 6; n' is 1, 2 or 3; n" > n; LZ<sub>n'+1</sub>, LZ<sub>n'+2</sub>, LZ<sub>n'+3</sub>, LZ<sub>n'-1</sub>, and LZ<sub>n'-2</sub> are application zones for liquids intended for transport of Reactant\* or other reactant or buffer without reactant.

24 (Previously Presented): The device according to claim 18, wherein the zones LZ<sub>m</sub> . LZ<sub>n</sub> . LZ<sub>1</sub> have zone spacers between each other.

25 (Previously Presented): The device according to claim 18, wherein at least one of the zones  $LZ_m \dots LZ_n \dots LZ_1$  comprises a pad or material layer applied on the flow matrix.

26 (Previously Presented): The device according to claim 18, wherein at least one reactant, other than Reactant\*, is pre-deposited in an application zone  $LZ_n^*R$  for liquid intended for transport of the reactant.

27 (Previously Presented): The device according to claim 18, wherein  $2 \leq m \leq 6$  and  $n'$  for the application zone for sample ( $LZ_nS$ ) is 1, 2 or 3.

28 (Previously Presented): The device according to claim 18, wherein the detection zone DZ comprises firmly anchored Reactant I, and a reactant to which Reactant I exhibits biospecific affinity optionally is pre-deposited in  $LZ_nS$  or in an application zone downstream of  $LZ_nS$ .

29 (Previously Presented): The device according to claim 18, wherein the flow matrix comprises at least one calibrator zone CZ, in which a calibrator or a binder for the calibrator is firmly anchored in the matrix.

30 (Previously Presented): The device according to claim 29, wherein the calibrator zone or zones (CZ) have a binder for the calibrator firmly anchored in the matrix, and calibrator optionally is pre-deposited in the matrix upstream of the calibrator zone or zones.

31 (Previously Presented): The device according to claim 18, wherein the device is intended for diagnosing allergy or autoimmune disease.

32 (Previously Presented): A test kit, comprising (i) a device according to claim 18, and (ii) Reactant\*.

33 (Previously Presented): The test kit according to claim 32, wherein the kit additionally comprises (iii) a calibrator when a binder for the calibrator is firmly anchored in the matrix.

34 (Previously Presented): The method according to claim 10, wherein each zone spacer comprises a strip attached to the flow matrix.

35 (Previously Presented): The device according to claim 24, wherein each zone spacer comprises a strip attached to the flow matrix.